

Insulating Biomaterials N01-NS-2-2347

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Neural Prosthesis Program



InnerSea Technology

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The goal of the Insulating Biomaterials work is to identify and evaluate materials, coatings, and assembly techniques suitable for protection of integrated circuit devices being considered for neural prosthetic applications.

New Instrumentation System

With the successful testing of the prototype quad charge integrator approach reported on in Progress Report 4, the control and data handling system approach was further developed. The goal was to develop a system where a unique test device code could be permanently connected to a particular device under test that could then be recognized by the data acquisition system. In addition, calibration information would be maintained in a small memory with each local quad integrator board. After a variety of approaches were investigated, an SPI bus architecture was chosen. The SPI bus is commonly implemented in inexpensive PIC microcontrollers. By including a microcontroller on each instrumentation card, control information could be passed from a central controller to each peripheral test device, and data from many peripheral test devices could be passed to a central controller. The central controllers would then communicate with the data acquisition computer by USB or Ethernet. Once implemented in this manner, test devices would be operating autonomously, passing data to the central computer when it was ready which would greatly speed the testing. More importantly, data integrity will be insured because of the self-testing features and permanently ID tags associated with each device under test.

Peel Testing

Considerable effort has been placed on refinement of the “Peel Test” to reduce the observed but unexplained high variability from sample to sample. As reported earlier, all possible environmental, silicone mixing, cure cycle, and test surface issues have been explored to no avail. Recently, microbubbles were identified at the surface interface and a new silicone formulation was specified to ensure that bubbling could be controlled. In addition, to guard against the



possibility that the hand-cut starting separation between the fiberglass tape and test surface was not a contributing variable, an attempt at identifying a narrow, thin release tape was made. An example of a test device with release tapes in place is shown in Figure 1. An example peel of a test slide with hand-cut Teflon release tape is shown in **Figure 2**. The simplest approach, that yielded the most consistent results, was a 1 mil thick Kapton tape with a silicone pressure sensitive adhesive. A vendor was identified that would provide a uniform batch of 1.5mm wide Kapton tape suitable for this application.

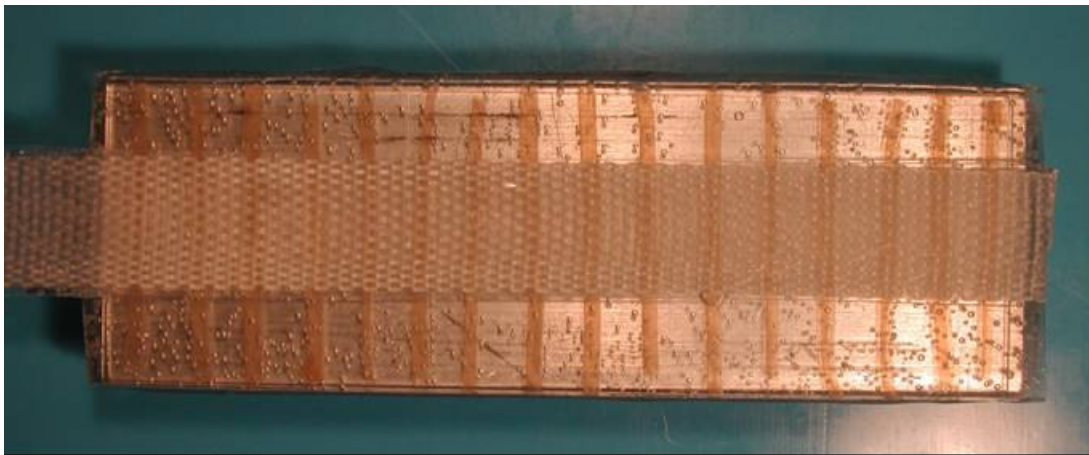


Figure 1: Example Peel Test Device with intermittent release tape stripes to ensure that peel forces are concentrated at the surface interface at the beginning of each peel test.

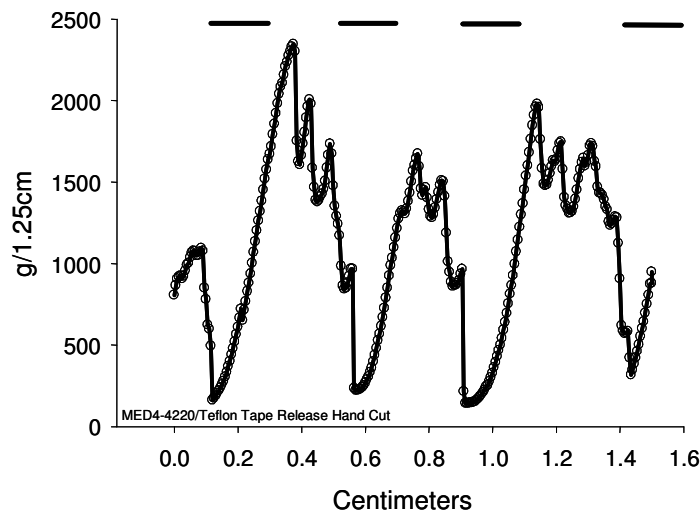


Figure 2: Example of release tape peel test device (Teflon release tape used in this example) showing intermittent release followed by peel from glass surface. Variability is reduced, but still significant.



CVD Synthesis of New Materials

The following is excerpted from Shannan O'Shaughnessy's recent thesis proposal, and reports on recent work and a research plan for future work on development of a flexible, insulating polymer coating suitable for micromachined integrated electrode arrays.

Proposed Work

Work on this thesis will focus on two areas: deposition of a flexible, insulating polymer coating by CVD, and surface functionalization of this coating to increase its biocompatibility. As success of the surface functionalization work is necessarily dependent on the development of a good coating to functionalize, primary efforts will be directed in this area. As noted, previous work within the Gleason group has achieved insulating polymeric coatings by CVD, but without the required flexibility for use in coating wires. It has also been previously demonstrated that coatings generated utilizing initiated organosilicon precursors (specifically V_3D_3) become more flexible as filament temperature is reduced (see Figure 4-1 below). However, previous film could not be made sufficiently flexible as the chosen radical initiator (PFOSF) required filament temperatures in excess of 350°C [2].

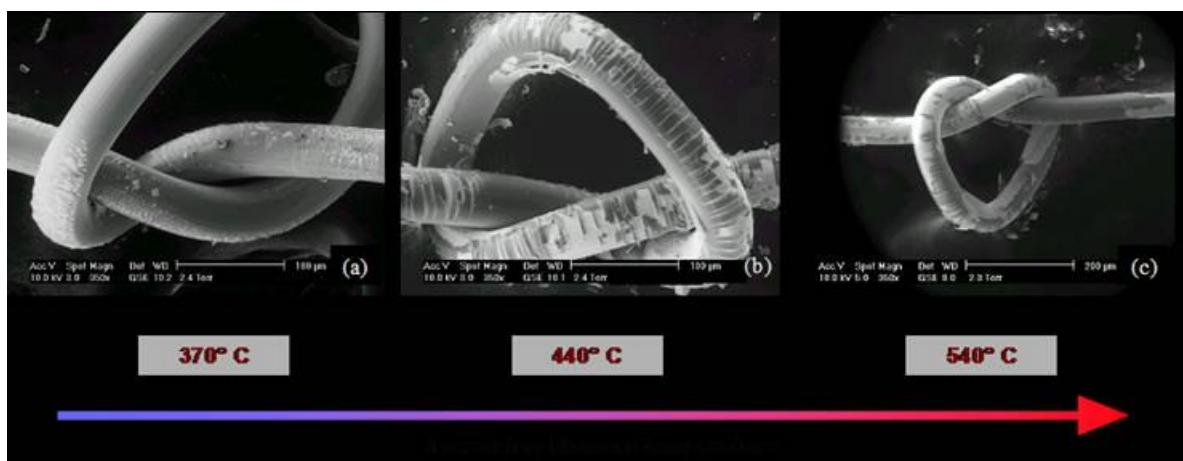


Figure 4-1: Effect of Filament Temperature on Film Flexibility[2]



In order to avoid this previous pitfall, research for this thesis will utilize radical initiators which activate polymerization at much lower filament temperatures. Specifically, work will focus on initiators from the peroxide family. Peroxides are often used as radical generating species for initiation of vinyl polymerization in solution. Previous work within the Gleason Group [46] has shown that tertbutylperoxide will decompose readily into butoxy radicals at temperatures between 180-220°C (see Figure 4-2). This is unsurprising as it is known that the same reaction will occur in solution at temperatures as low as 150°C [47]. At these temperatures, utilizing V_3D_3 as the organosilicon precursor, it should be possible to create a networked polymer film which is flexible enough for the application. In addition, the multiple vinyl moieties present on the V_3D_3 monomer should create enough crosslinking to provide the material with sufficient mechanical toughness. The level of crosslinking can be tuned by control of the precursor to initiator ratio.

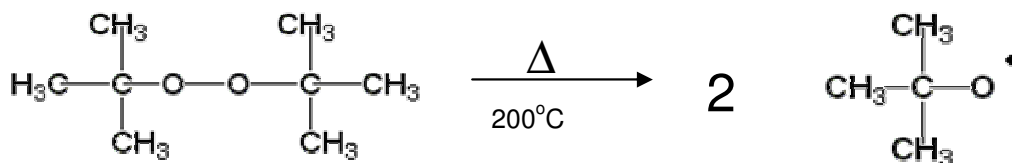


Figure 4-2: Use of Tertbutylperoxide as a Radical Generating Initiator

In addition to V_3D_3 , other organosilicon precursors possessing fewer vinyl functional groups will be investigated. One good candidate precursor is vinylmethylbis(trimethylsiloxy)silane (figure 4-3a). Possessing only one vinyl group, this precursor should form linear chains when radically polymerized. This will guarantee film flexibility, but does not provide for mechanical toughness. Required toughness will be introduced through use of a crosslinking agent such as 1,5-Divinylhexamethyltrisiloxane (Figure 4-3b). By controlling the ratio of these two precursors, optimization of film flexibility vs. toughness should be achievable.

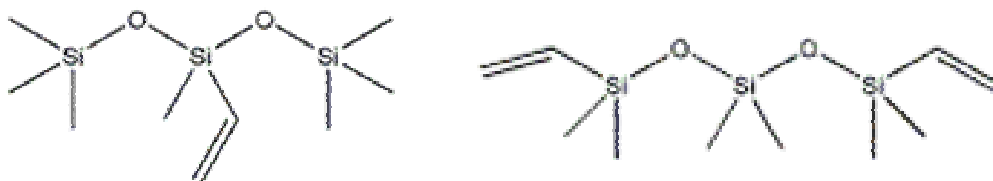


Figure 4-3a &b: Vinylmethylbis(trimethylsiloxy)silane and 1,5-Divinylhexamethyltrisiloxane

Once the primary goal of synthesizing a novel polymeric film with bulk resistivity on the order of $1e14$ Ohm-cm and the required flexibility for the application has been achieved, research will then be refocused on the surface modification of the designed material in order to improve biocompatibility. Specifically, the goal will be improved adhesion of neurons to the material and growth of said neurons in order to allow for better recording over time from neural electrodes coated with the polymer. It should be noted, however, that continued testing of the polymeric film for long term stability in a saline environment will continue even while surface modification work is taking place. This will insure that the base insulating coating possesses the required chemical stability for long term implantation.

Multiple bioactive molecules suitable for surface modification of the coating have been identified in the literature, including poly-L-lysine (PLL), laminin, fibronectin, and active fibronectin fragments such as RGD (made up of a bioactive combination of arginine, glycine, and aspartic acid) [34-38, 43, 48-52]. This research will concentrate primarily on the use of PLL as it can be readily covalently bonded to the polymer coating. PLL possesses many amide which can be readily utilized to bond with a polymer (see Figure 4-4). Both sets of proposed precursors possess vinyl moieties (through which polymerization will be taking place). In addition, it is anticipated that the resulting polymer will possess these some of these groups, unreacted, on its surface. These vinyl groups will be utilized for attachment of PLL. This can be done one of two ways: either through direct conversion to a reactive ester or carboxylic acid group and



attachment, or through addition of another polymer chain at to the vinyl moiety which itself possesses multiple reactive groups for attachment. In the first approach, an acid, such as HBr or HCl, would be used attack the vinyl bond through electrophilic addition. This would result in the chlorination (or bromination) of the carbon directly attached to the bulk polymer film. This halide activated site would then be reacted with an alcohol containing a carboxylic acid moiety. Finally, the PLL would be attached through nucleophilic reaction of the PLL amide with the carboxylic acid. This final step would be catalyzed by the use of carbodiimide to ensure attachment reactions occur at all acid groups. In the second methodology, polyacrylamide (PAM) would be polymerized in a solution contacting the deposited coating. As PAM polymerization is through a vinyl moiety, some PAM chains should polymerize across the vinyl bonds present on the surface of the coating. This has previously been demonstrated by Murthy et al. [2]. Attached PAM chains would then be hydrolyzed to polyacrylic acid, and then functionalized with PLL utilizing the same carbodiimide catalyzed nucleophilic addition detailed above. The second methodology has the obvious advantage of allowing multiple PLL attachments at each vinyl site. Both attachment chemistries would take place in aqueous solution to avoid contacting the polymer with any solvents. Surface modified films will then be analyzed as described in Section 3.0 for increased biocompatibility with neurons.

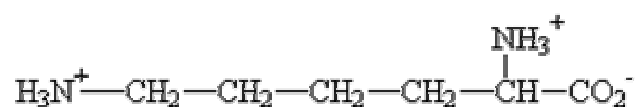


Figure 4-4: L-Lysine in Its Protonated Form

Lastly, creation of a copolymer will be explored. The purpose of the copolymerization will primarily be the introduction of an additional moiety (such as a methacrylic acid) for direct attachment of the PLL. Attachment would take place as above, through nucleophilic addition of the PLL amide group to the carboxylic acid present in the copolymer. This will avoid the necessity of multiple



solution phase reaction steps. In addition, the amount of PLL will be controllable through the ratio of the primary precursor to the 'attachment moiety' precursor. As the additional groups will not participate in the polymerization reaction, their only purpose will be for attachment, and their surface density should be controllable. In fact, it may be possible to introduce a gradation in the coating, only adding the 'attachment moiety' precursor near the end of deposition for optimal control of both surface and bulk material properties

Preliminary Results

As stated above, initial work will focus on the development of the polymeric coating itself. As such, depositions have been performed utilizing V_3D_3 as the primary precursor with radical initiation by tertbutylperoxide. Depositions were performed at ~300mTorr pressure, with a substrate stage temperature of 25°C. Filaments of both Nichrome (85% Nickel, 15% Chromium) and Constantan (55% Copper, 45% Nickel) were utilized for thermal activation of the initiator. The filament array was arranged at a height of 3cm above the stage, and filament temperature was varied between 200-275°C. In addition to varying filament temperature, the ratio of precursor to initiator was also varied between 50:1 and 1:1. Depositions were made on both flat silicon and 50 μ m platinum wire substrates. It should be noted that while the silicon flat can be assumed to be at ~25°C as it is in direct contact with the stage, the platinum wire can not be assumed to maintain constant temperature as it is held significantly above the stage for coating.

Results from these depositions were mostly positive. Coatings on both wires and flats were extremely conformal. In addition, FTIR analysis of the samples (Figure 5-1) indicated that polymerization had taken place as planned, with the polymer backbone formed through opening of the vinyl moiety as in Figure 5-2.

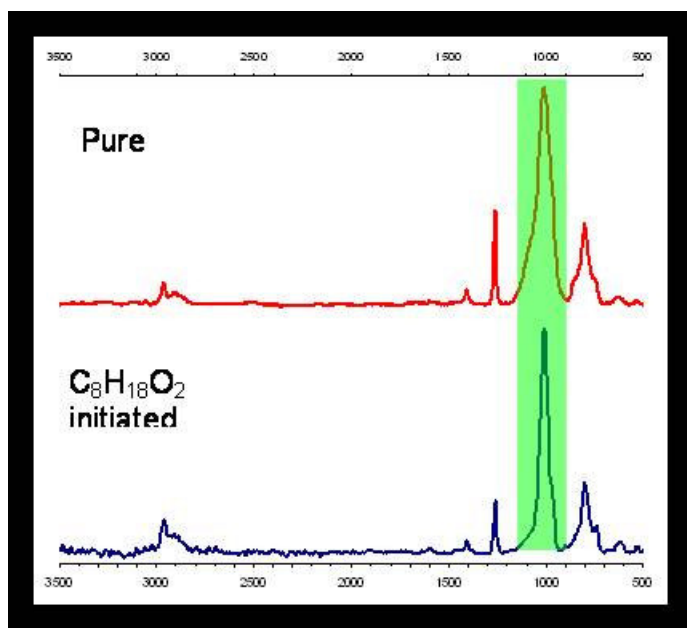


Figure 5-1: FTIR Spectra of Pure V_3D_3 and that of Tertbutylperoxide Initiated V_3D_3

In addition, the highlighted area of the FTIR spectra indicates that the siloxane rings of the V_3D_3 precursor have been retained with no ring opening side reactions (as discussed in Section 2.2). The highlighted peak (at ~ 1025 wavenumbers) represents the Si-O-Si stretch in the organosilicon ring of V_3D_3 . The existence of this peak as a singlet demonstrates that all Si-O-Si bonds still exist in the form of rings. Had any ring opening occurred during deposition, the peak would show doublet character, with some of its intensity shifted ~ 50 wavenumbers higher. Finally, wire coatings show excellent flexibility, with no visible cracking or loss of continuity even when knotted (see Figure 5-3a). It should be noted that the knot in Figure 5-3a is only $\sim 125\ \mu\text{m}$ in diameter, at least 4X smaller than those attempted in previous work.

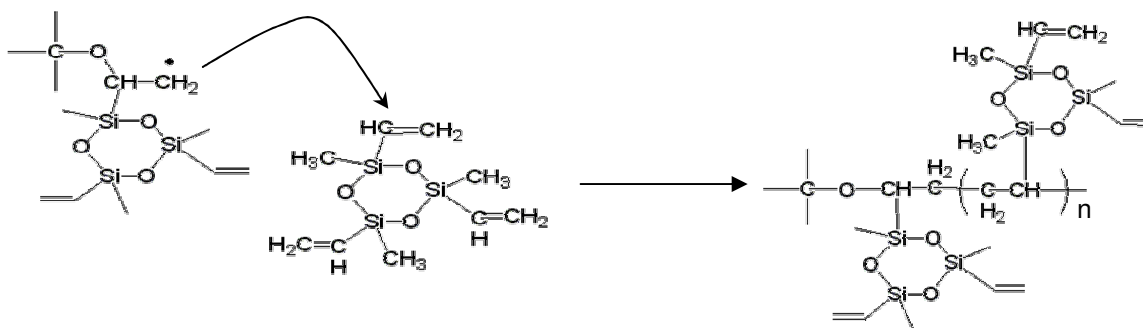


Figure 5-2: Polymerization of V_3D_3 by Tertbutylperoxide Initiation

Unfortunately, these coatings also have certain problems. The primary issue is that they experience significant thickness loss upon reactor blowdown at the end of deposition. As this thickness loss does not continue after removal of the sample from the reactor, it is theorized that it is due to vaporization of low molecular weight polymer that occurs when the pressure is reduced. This vaporization unfortunately results in small flaws in the coating (for thin films) and can create large bubbles in thicker films. In addition, the vaporization seems to flaw the coating enough as to destroy its electrical resistivity. This is visually demonstrated in Figure 5-3b, a coated wire loop which has been resistively tested in saline solution. Further work will concentrate on manipulation of deposition parameters to increase polymer molecular weight. There are multiple strategies which may help in increasing the polymer molecular weight. Most basic would be the increase of residence time in the reactor by decreasing overall precursor flow rates. In addition, an increase of the precursor to initiator ratio may help in avoiding early termination of chains (though this will require additional hardware for initiator control). Another approach which may help to increase molecular weight would be altering the temperature of reaction. While it is unclear whether the polymerization is taking place in the gas phase (with complete polymer molecules depositing on the wafer) or on the surface of the substrate (with initiated molecules depositing and growing polymer there), both the filament temperature and the substrate temperature are controllable. The literature shows many examples of polymerizations where the rate of propagation has a much higher activation energy than the rate of termination [53, 54]. As this



is most likely the case in the considered polymerization, an increase in the appropriate reaction temperature should favor higher molecular weight polymer. A better understanding of the kinetics of the polymerization, and thus of the drivers for higher or lower polymer chain length, could be gained through a temperature series of experiments at high initiator ratios (where the reaction is no longer dependent on initiator concentration). Lastly, an increase in substrate temperature or a decrease in reactor pressure would limit the ability of low molecular weight species to adsorb on the substrate, and would therefore assist in decreasing the loss of film thickness upon reactor blowdown.

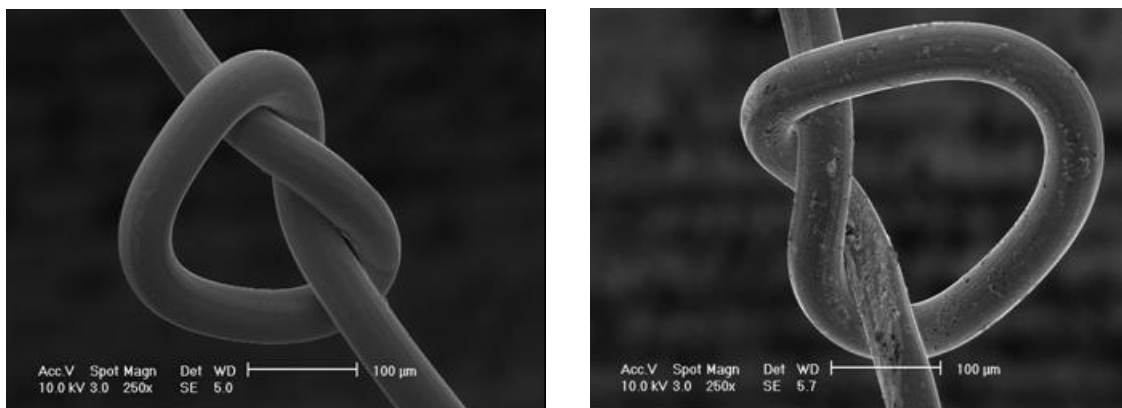


Figure 5-3a & b: Coated 50 μ m wire untested (left) and after resistivity testing (right)

6.0 Safety

There are four areas of safety concern associated with this project. The first, and most important, is the safe handling of precursor chemicals. While the organosilicon precursors are fairly benign, peroxide initiators can be both toxic and, if not correctly handled, explosive. In addition, most experimental compounds, such as V3D3, have not been assessed for long term exposure risk. Therefore, utmost care will be taken in handling and storage of chemical precursors, including use of proper PPE (safety glasses, gloves, apron and respirator when appropriate) and refrigeration of stored peroxides.



Additional safety concerns associated with the project include use of reactor and vacuum pumps, use of analysis equipment, and handling of biological samples for biocompatibility testing. Appropriate safety precautions will be taken when utilizing the CVD reactor including allowing adequate time for precursor gasses to vent and the filament to cool before opening the reactor after deposition. All analysis equipment will be used as directed, and proper PPE will be worn while handling biological samples (gloves and safety glasses at a minimum).

7.0 Proposed Schedule

	2003	2004			2005			2006		
	Fall	Spring	Sum	Fall	Spring	Sum	Fall	Spring	Sum	Fall
Development and Characterization of V3D3 based Film										
Development and Characterization of Alternative Precursor Film										
Surface Functionalization										
Biopassivation and Biocompatibility Testing										
Thesis										